

Photosynthesis

Revision notes • IB Biology C1.3: energy transformation, pigments, chromatography, spectra, limiting factors, CO₂ enrichment, HL light reactions and the Calvin cycle.

For IB Diploma Biology, Standard Level and Higher Level (first assessment 2025). **HL only** is additional higher level content, for HL students only.

Photosynthesis Spec C1.3

Photosynthesis: energy, carbon dioxide, water and oxygen Spec C1.3.1, C1.3.2, C1.3.3

Photosynthesis involves the **transformation of light energy to chemical energy**. Light energy absorbed by pigments is used to make carbon compounds, and the energy is then held in the chemical bonds of those compounds. This energy transformation supplies most of the chemical energy needed for life processes in ecosystems: producers use it directly, and consumers and decomposers obtain it by feeding.

Carbon dioxide, water and glucose

Carbon dioxide is converted to glucose using **hydrogen obtained by splitting water**. Carbon dioxide provides the carbon and oxygen atoms of glucose, but glucose also contains hydrogen, which must come from somewhere: it comes from water molecules, split using energy from light.



Oxygen as a by-product

When water is split, the hydrogen is used and the oxygen is released as a **by-product**. So **the oxygen produced by photosynthesis comes from the splitting of water**, not from carbon dioxide. Oxygen-producing photosynthesis happens in **plants, algae and cyanobacteria**.

Common mistake: Do not write that the oxygen released comes from carbon dioxide. It comes from water; the oxygen atoms of CO₂ end up in glucose and water.

Exam tip: Linking question: the consequences of photosynthesis for ecosystems include supplying chemical energy to food chains and removing carbon dioxide from the atmosphere.

Separating photosynthetic pigments by chromatography Spec C1.3.4

Leaves contain several photosynthetic pigments. They can be separated by **chromatography**, using **paper** or a **thin-layer chromatography (TLC)** plate. Each pigment travels a different distance because it differs in how soluble it is in the solvent and how strongly it is attracted to the stationary phase.

1. Grind leaf tissue with a little sand and propanone (or another suitable solvent) to extract the pigments.
2. Draw a pencil origin line about 1 cm from the bottom of the paper or plate.
3. Apply a small, concentrated spot of extract to the origin, letting it dry between applications.
4. Stand the paper or plate in a small depth of running solvent, below the origin, in a covered container.
5. Remove it when the solvent is near the top and immediately mark the solvent front with pencil.

6 . Mark the centre of each pigment spot, measure distances from the origin and calculate Rf values.

$$\text{Rf} = \text{distance moved by pigment} \div \text{distance moved by solvent front}$$

Rf values are always between 0 and 1 and have no units. Pigments are identified by their **colour** and by comparing their **Rf values** with reference values for the same solvent and stationary phase.

Common photosynthetic pigments and their colours

Pigment	Colour on the chromatogram
Carotene	Orange-yellow (usually travels furthest)
Xanthophylls	Yellow
Chlorophyll a	Blue-green
Chlorophyll b	Yellow-green

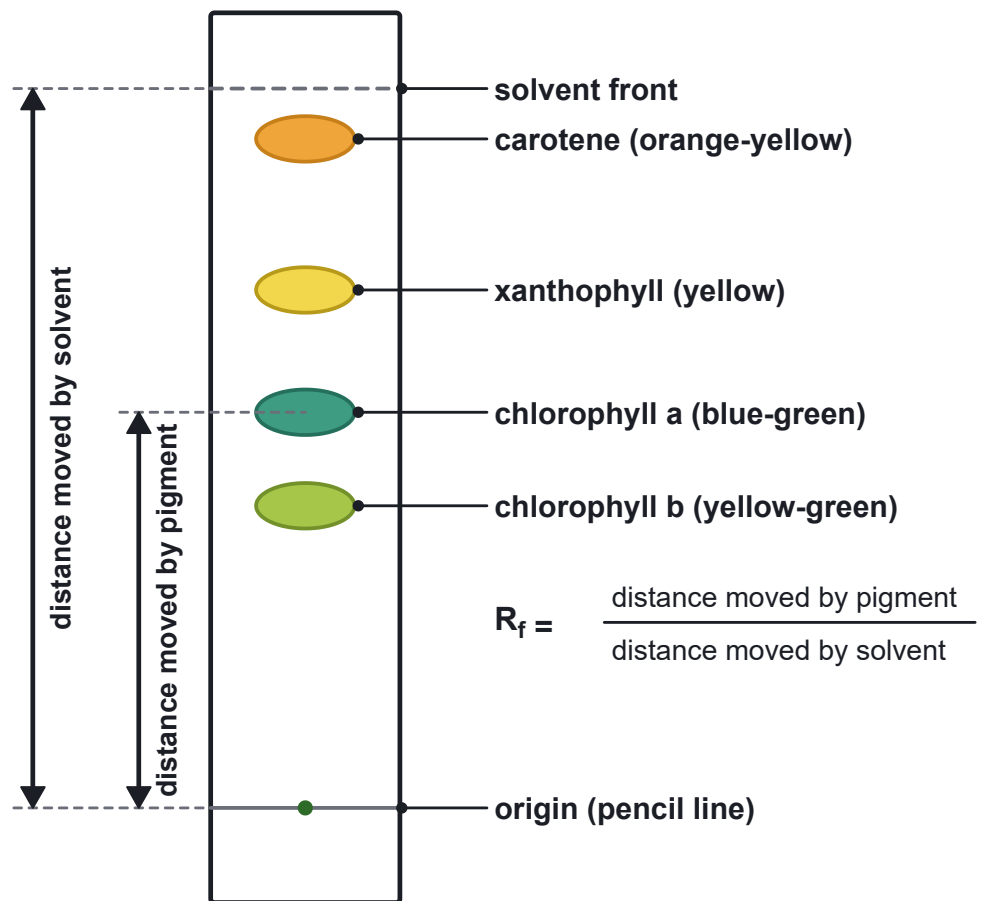
Calculating an Rf value

On a chromatogram the solvent front moved 8.0 cm from the origin. A blue-green spot moved 4.4 cm. Calculate its Rf value.

- 1 . $\text{Rf} = \text{distance moved by pigment} \div \text{distance moved by solvent front}$.
- 2 . $\text{Rf} = 4.4 \div 8.0 = 0.55$.

Answer: $\text{Rf} = 0.55$ (no units). With its blue-green colour this is likely chlorophyll a; confirm with reference values for the same method.

Practical skill: Use a pencil for the origin (ink would separate too), keep the origin above the solvent, and measure to the centre of each spot. Rf values depend on the solvent and medium, so only compare with references from the same method.



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A typical chromatogram of leaf pigments (illustrative positions; order and R_f values depend on the solvent and medium). $R_f = \frac{\text{pigment distance}}{\text{solvent distance}}$.

Absorption of light, absorption spectra and action spectra Spec C1.3.5, C1.3.6

Absorption of specific wavelengths

When a pigment molecule absorbs light, the energy **excites an electron** within the molecule, raising it to a higher energy level. This is the first step in transforming light energy to chemical energy, because the excited electron can be passed on to other molecules.

Only some wavelengths are absorbed. Electrons in a pigment can only occupy certain energy levels, so only photons whose energy matches the gap between levels can be absorbed. Other wavelengths are reflected or transmitted. **Chlorophyll** absorbs red and blue light most strongly and reflects green light, which is why leaves look green. Accessory pigments such as carotenoids absorb other wavelengths, mainly in the blue-green region.

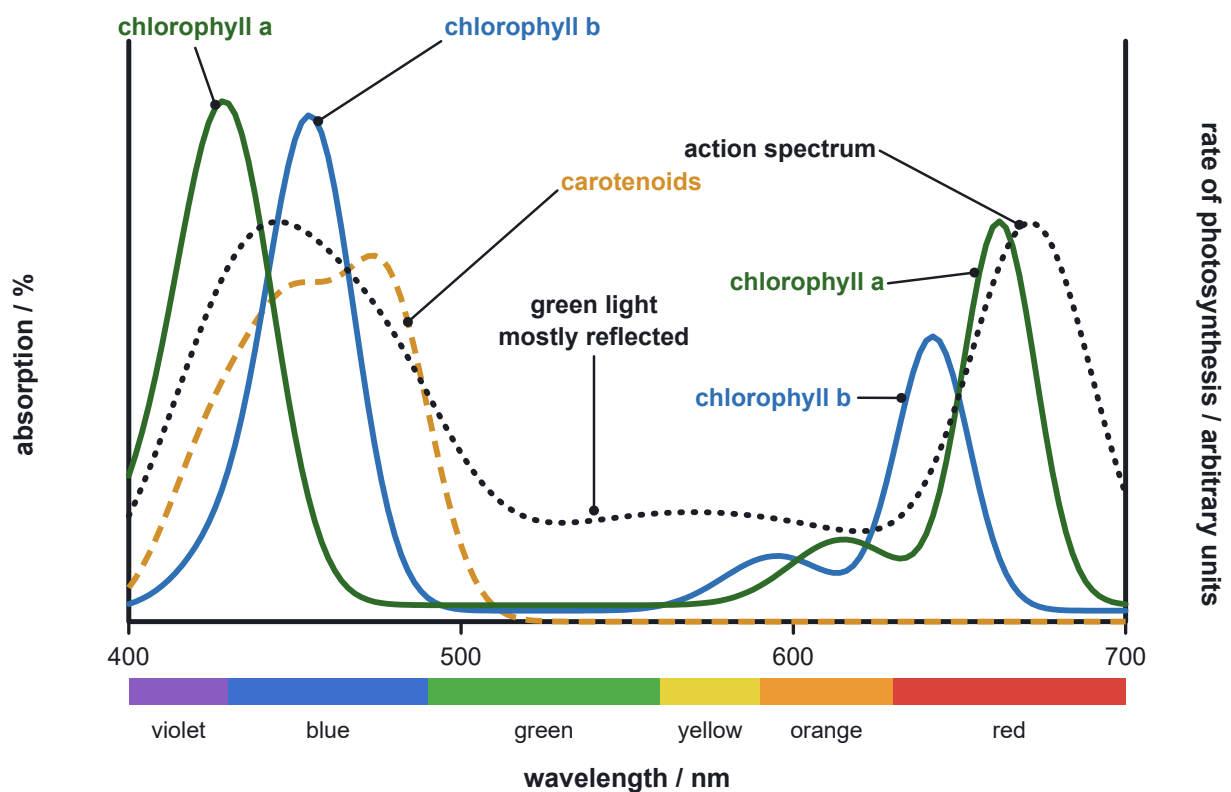
Absorption spectra and action spectra

Absorption spectrum	A graph showing the percentage of light absorbed by a pigment (or pigments) at each wavelength.
Action spectrum	A graph showing the rate of photosynthesis at each wavelength of light.

Both graphs have **wavelength** (in nm, from about 400 to 700 nm) on the horizontal axis, which should also show the **colours** of light: violet and blue at the short-wavelength end, then green, yellow, orange and red.

Similarities and differences of absorption and action spectra		
	Absorption spectrum	Action spectrum
Vertical axis	% of light absorbed	Rate of photosynthesis
Peaks	Blue and red for chlorophyll	Also blue and red
Green region	Low absorption by chlorophyll	Rate low but not zero, because accessory pigments absorb some light and some green light is absorbed as it passes through the leaf
How obtained	Shine each wavelength through a pigment extract and measure absorbance	Measure rate of oxygen production or carbon dioxide consumption under each wavelength

The **similar shape** of the two graphs is evidence that the light absorbed by these pigments is the light used in photosynthesis.



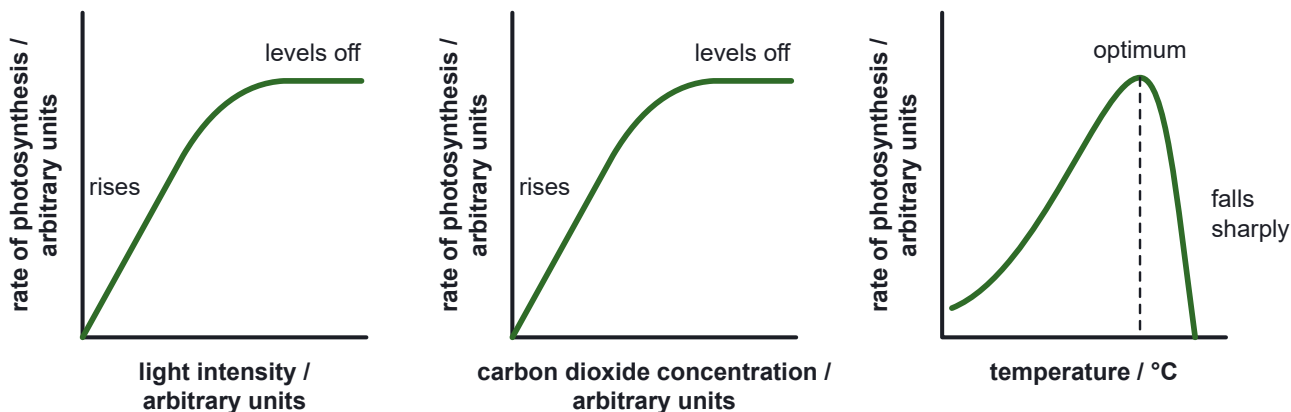
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Illustrative curves: the action spectrum **matches the absorption spectra**, with peaks in blue and red light; green light is mostly reflected.

Maths skill: To make an action spectrum, calculate the rate at each wavelength (for example volume of oxygen $\div$ time, or CO_2 uptake $\div$ time), then plot rate against wavelength with points joined by a smooth curve or straight lines.

Investigating limiting factors on photosynthesis Spec C1.3.7

The rate of photosynthesis depends on several factors. At any moment, the **limiting factor** is the one that is furthest from its optimum: increasing it raises the rate, while increasing the others does not.



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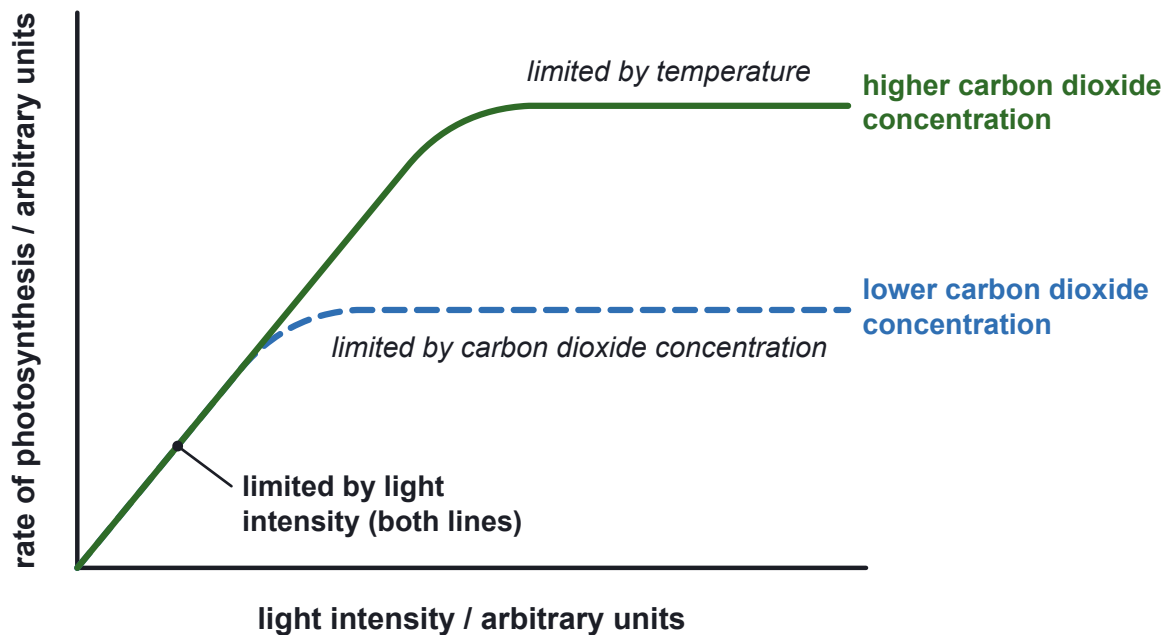
Light intensity and carbon dioxide concentration: the rate **rises, then levels off** when another factor limits. Temperature: the rate rises to an optimum, then falls as enzymes denature.

Techniques for varying each factor

How to vary each limiting factor		
Independent variable	How to vary it	Notes
Light intensity	Change the distance of a lamp from the plant, or use filters or a dimmable LED	Light intensity falls with distance (roughly proportional to $1 \div \text{distance}^2$); use an LED or a heat shield so temperature does not change
Carbon dioxide concentration	Use different concentrations of sodium hydrogencarbonate solution around aquatic plants or leaf discs	Hydrogencarbonate releases CO_2 in water
Temperature	Place the plant in water baths at different temperatures	Allow time to equilibrate; check with a thermometer

Measuring the rate

- Count bubbles or collect the volume of oxygen released by an aquatic plant such as *Elodea* or *Cabomba* per minute.
- Time how long leaf discs (with air removed so they sink) take to float as oxygen builds up.
- Measure carbon dioxide uptake with a sensor and data logger, or by the colour change of a hydrogencarbonate indicator.



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Where each line levels off, light is no longer limiting: the **factor in shortest supply** sets the plateau.

Practical skill: Identify the independent variable (the factor you change) and the dependent variable (the rate, for example oxygen per minute). Keep the other two factors constant and repeat each condition.

Exam tip: A hypothesis is a provisional explanation that needs repeated testing. You can suggest one from theory before the experiment (for example, 'rate rises with light intensity because more light energy is absorbed by chlorophyll') or from results after it.

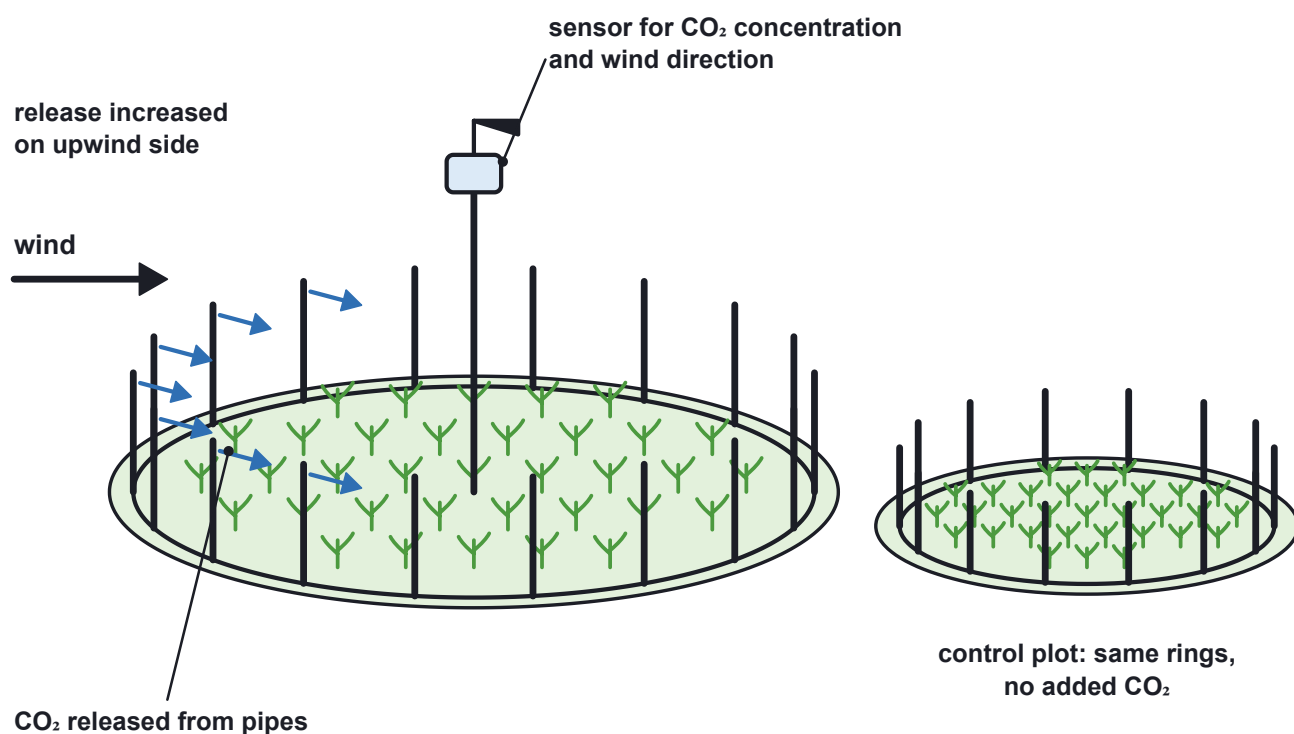
Carbon dioxide enrichment experiments **Spec C1.3.8**

Atmospheric carbon dioxide concentration is rising. **Carbon dioxide enrichment experiments** grow plants at concentrations predicted for the future to find out how rates of photosynthesis and plant growth may change.

Two types of CO ₂ enrichment experiment		
	Enclosed greenhouse experiments	Free-air CO ₂ enrichment (FACE)
Set-up	Plants grown in closed glasshouses or chambers with CO ₂ added to the air	Rings of pipes release CO ₂ around plots in a field or natural ecosystem; sensors adjust the release to hold a target concentration
Control of variables	Easier: temperature, light, water and minerals can be controlled	Harder: weather, soil and pests vary, but affect treated and control plots similarly
How realistic	Less natural: enclosure changes temperature, humidity and wind	More realistic: plants grow in field conditions with other species
Control	Identical greenhouse at current CO ₂	Plots with the same ring structure but no added CO ₂

Higher carbon dioxide generally increases the rate of photosynthesis and growth, at least for a time. However, several FACE studies of crops found **smaller increases** in growth and yield than enclosed experiments had predicted (the size of the gap is still debated). In the field, other factors such as water, temperature or soil nitrogen can become limiting, and the effect of extra CO₂ can shrink over the years.

Practical skill: Be able to identify a controlled variable, such as plant species, light intensity in a greenhouse, or plot size in a FACE experiment. Field experiments are less controlled than laboratory ones, but some questions can only be answered in natural ecosystems.



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A FACE experiment: pipes release CO₂ around a field plot to hold a target concentration; control plots have the same rings but no added CO₂.

Photosystems and the photolysis of water Spec C1.3.9, C1.3.10, C1.3.11

HL only

Photosystems

A **photosystem** is a molecular array of chlorophyll and accessory pigments, held in position by proteins, with a **special chlorophyll** at the **reaction centre**. Photosystems are always located in **membranes**: in the thylakoids of chloroplasts in photosynthetic eukaryotes, and in membranes inside cyanobacteria.

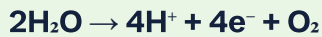
Pigment molecules around the reaction centre absorb light and pass the energy from molecule to molecule until it reaches the reaction centre chlorophyll. There an electron is excited so strongly that it is **emitted** and passed to an electron acceptor. There are two types: photosystem II and photosystem I.

Advantages of a structured array

- Different pigments absorb different wavelengths, so the array captures energy from a wider range of light.
- Many pigment molecules capture far more photons than one, and all their energy is funnelled to one reaction centre.
- The reaction centre is positioned next to an electron acceptor, so the excited electron is passed on instead of falling back and losing its energy as heat or light.
- A single molecule of chlorophyll or any other pigment would not be able to perform any part of photosynthesis on its own.

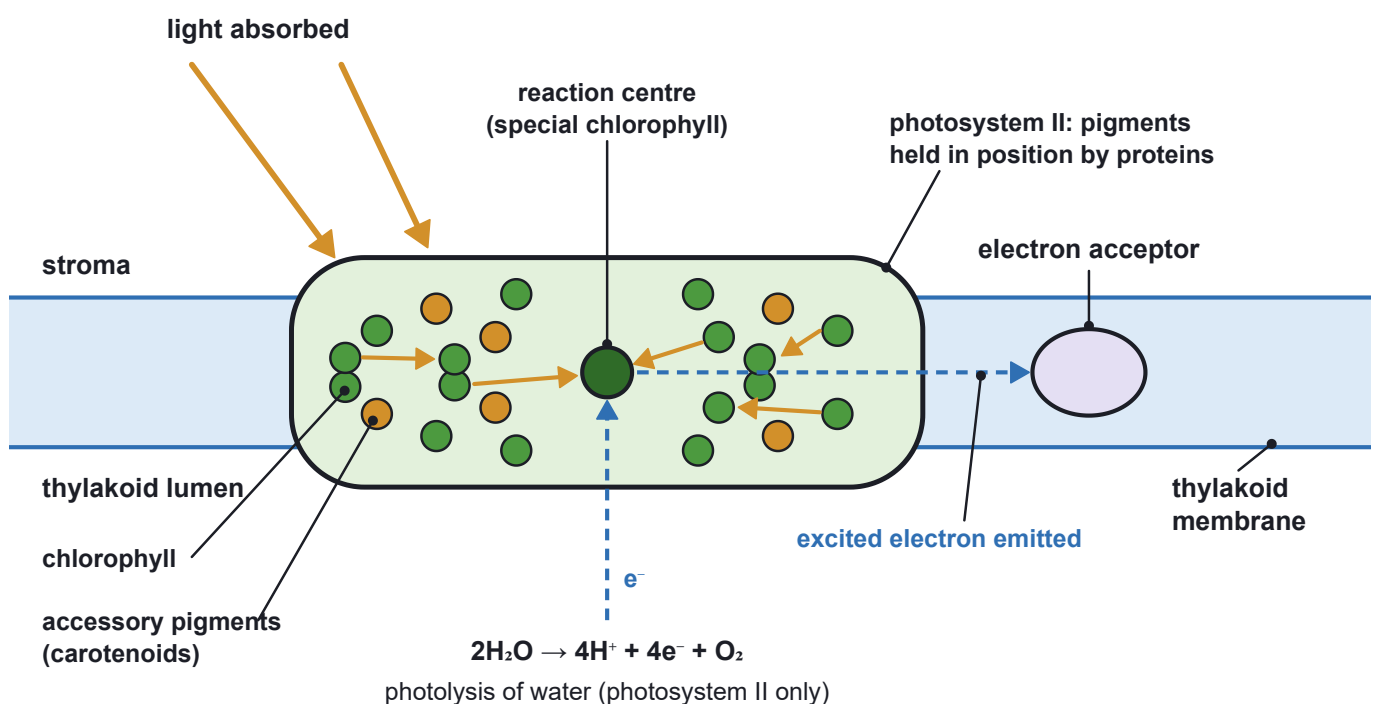
Photolysis of water in photosystem II

Photosystem II loses electrons from its reaction centre. They are replaced by **photolysis**: the splitting of water, catalysed by an enzyme complex in photosystem II on the thylakoid lumen side of the membrane.



The **electrons** replace those emitted by photosystem II, and the **protons**, released into the thylakoid lumen, add to the proton gradient used to make ATP by chemiosmosis. Both are used in photosynthesis, but **oxygen is a waste product** and diffuses out.

The advent of oxygen generation by photolysis, in early cyanobacteria, had immense consequences for life and geological processes on Earth. Oxygen accumulated in the oceans and atmosphere, dissolved iron was oxidised and precipitated (forming banded iron rock deposits), an ozone layer formed, and aerobic respiration became possible, while many anaerobic organisms were poisoned.



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A photosystem funnels light energy from many pigments to the **reaction centre**, which emits an excited electron. In photosystem II, photolysis replaces it.

Chemiosmosis, reduction of NADP and thylakoids

Spec C1.3.12, C1.3.13, C1.3.14

HL only

ATP production by chemiosmosis

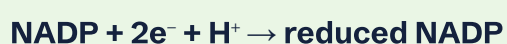
1. Excited electrons emitted by a photosystem pass along a **chain of electron carriers** in the thylakoid membrane.
2. Energy released as the electrons flow is used to **pump protons** from the stroma into the thylakoid lumen.
3. Protons (also released into the lumen by photolysis) build up a **proton gradient**: high concentration in the lumen, low in the stroma.
4. Protons diffuse back into the stroma through **ATP synthase**, which uses the energy to phosphorylate ADP, releasing ATP into the stroma. This is **chemiosmosis**.

Two ways of supplying electrons for ATP production

	Non-cyclic photophosphorylation	Cyclic photophosphorylation
Source of electrons	Photosystem II	Photosystem I
Path of electrons	PSII → carrier chain → PSI → NADP	PSI → carrier chain → back to PSI
Products	ATP, reduced NADP and oxygen	ATP only
Photolysis of water	Yes	No

Reduction of NADP by photosystem I

Photosystem I absorbs light and emits excited electrons. NADP is reduced by **accepting two electrons** that have come from photosystem I. It also **accepts a hydrogen ion from the stroma**, forming **reduced NADP**. Electrons lost from photosystem I are replaced by electrons arriving along the carrier chain from photosystem II.



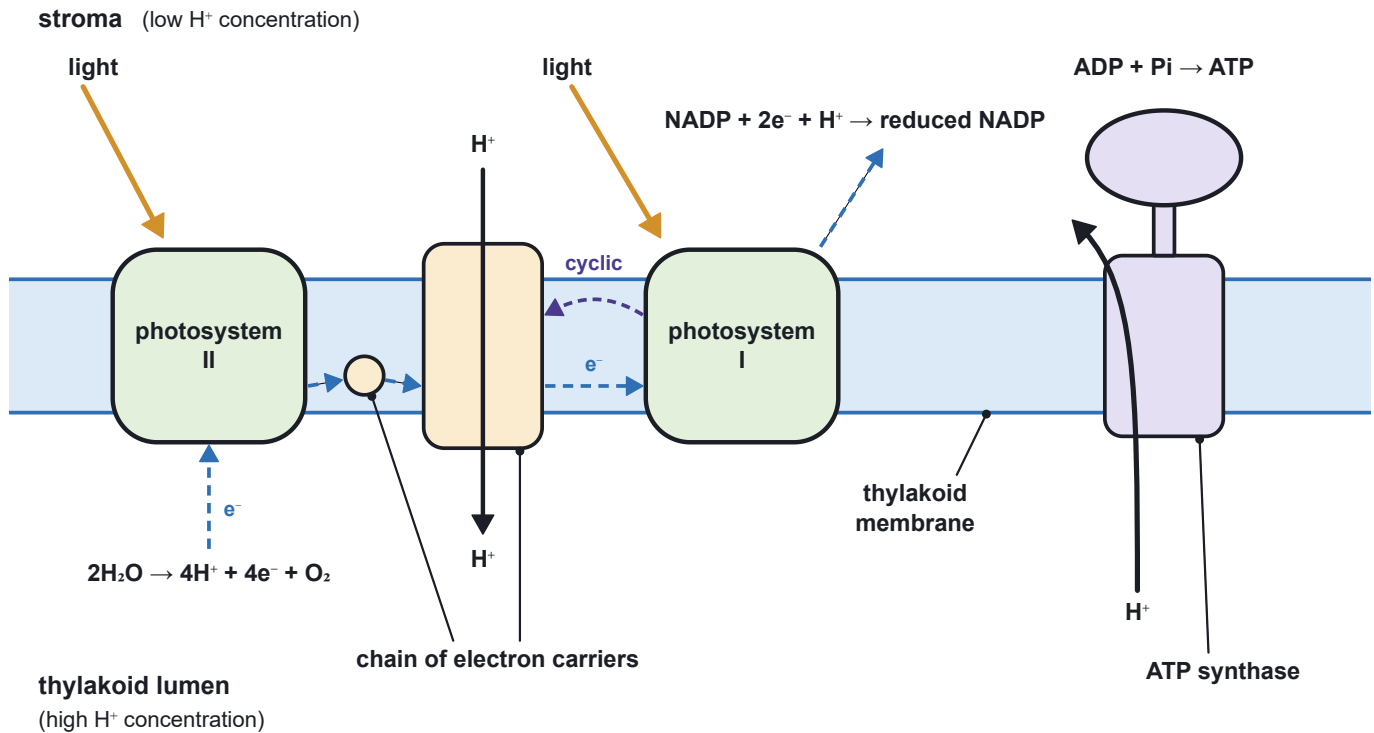
Exam tip: Pair the terms consistently: write 'NADP and reduced NADP' or 'NADP⁺ and NADPH', never a mixture such as 'NADP and NADPH'.

Thylakoids as systems for the light-dependent reactions

Where each light-dependent process happens in a thylakoid

Process	Location
Light absorption by photosystems	Thylakoid membrane
Photolysis of water	Photosystem II, on the lumen side of the membrane; protons released into the lumen
Proton pumping	Electron carriers in the membrane, from stroma into lumen
ATP synthesis by chemiosmosis	ATP synthase in the membrane; ATP released into the stroma
Reduction of NADP	Stroma side of photosystem I

The small volume of the thylakoid lumen means a steep proton gradient builds up quickly, and stacking thylakoids into grana gives a large membrane area for photosystems. ATP and reduced NADP are both released into the **stroma**, where the Calvin cycle uses them.



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The light-dependent reactions: electrons flow from **photosystem II to photosystem I to NADP**, building a proton gradient that drives ATP synthase.

The Calvin cycle: carbon fixation, triose phosphate and RuBP Spec C1.3.15, C1.3.16, C1.3.17

HL only

Carbon fixation by Rubisco

In the stroma, the enzyme **Rubisco** catalyses the combination of **carbon dioxide** with **RuBP** (ribulose biphosphate, 5C). The product splits immediately into two molecules of **glycerate 3-phosphate (GP)**, a 3C compound. This is carbon fixation: inorganic carbon becomes part of an organic molecule.



Rubisco is the **most abundant enzyme on Earth**. High concentrations of it are needed in the stroma because it works relatively slowly and is not effective in low carbon dioxide concentrations.

Synthesis of triose phosphate

Glycerate 3-phosphate is converted into **triose phosphate (TP)** using reduced NADP and ATP from the light-dependent reactions. Reduced NADP supplies the hydrogen (GP is reduced) and ATP supplies energy.

Regeneration of RuBP

Most triose phosphate is used to regenerate RuBP, using more ATP. You do not need the individual reactions, but **five molecules of triose phosphate are converted to three molecules of RuBP** ($5 \times 3C = 15C = 3 \times 5C$), allowing the Calvin cycle to continue.

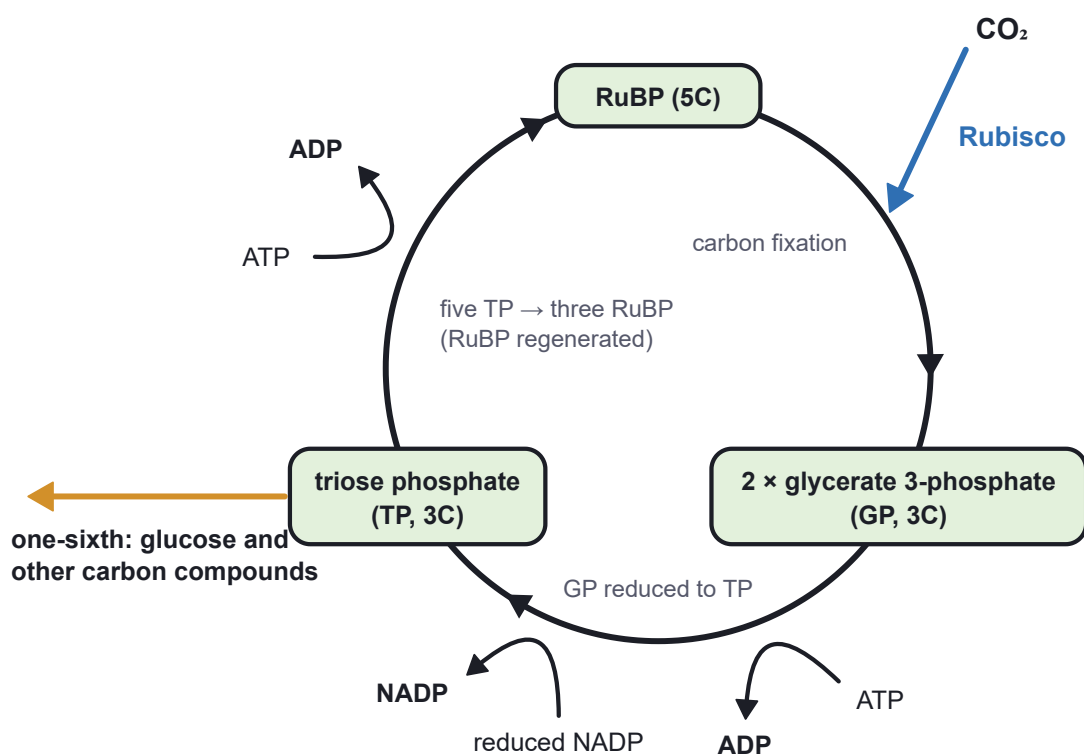
Why five-sixths of the triose phosphate is recycled

Show that if glucose is the product, five-sixths of the triose phosphate must be converted back to RuBP.

1. One glucose (6C) needs six carbon atoms, so 6 CO₂ must be fixed by 6 RuBP.
2. $6 \text{ RuBP} + 6 \text{ CO}_2 \rightarrow 12 \text{ GP} \rightarrow 12 \text{ TP}$ (36 carbon atoms).
3. To regenerate the 6 RuBP (30 carbon atoms), 10 TP are needed ($10 \times 3\text{C} = 30\text{C}$).
4. That leaves 2 TP (6C) to make one glucose.
5. 10 of 12 TP are recycled: $10 \div 12 = 5/6$.

Answer: Five-sixths of the TP is used to regenerate RuBP and one-sixth forms glucose.

stroma



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The Calvin cycle: carbon fixation by Rubisco, reduction of GP to TP, and regeneration of RuBP.

Exam tip: Use the IB names: RuBP, glycerate 3-phosphate (GP) and triose phosphate (TP). The intermediate between RuBP and GP is not required.

Making other carbon compounds and interdependence of the reactions

Spec C1.3.18, C1.3.19

HL only

Synthesis of other carbon compounds

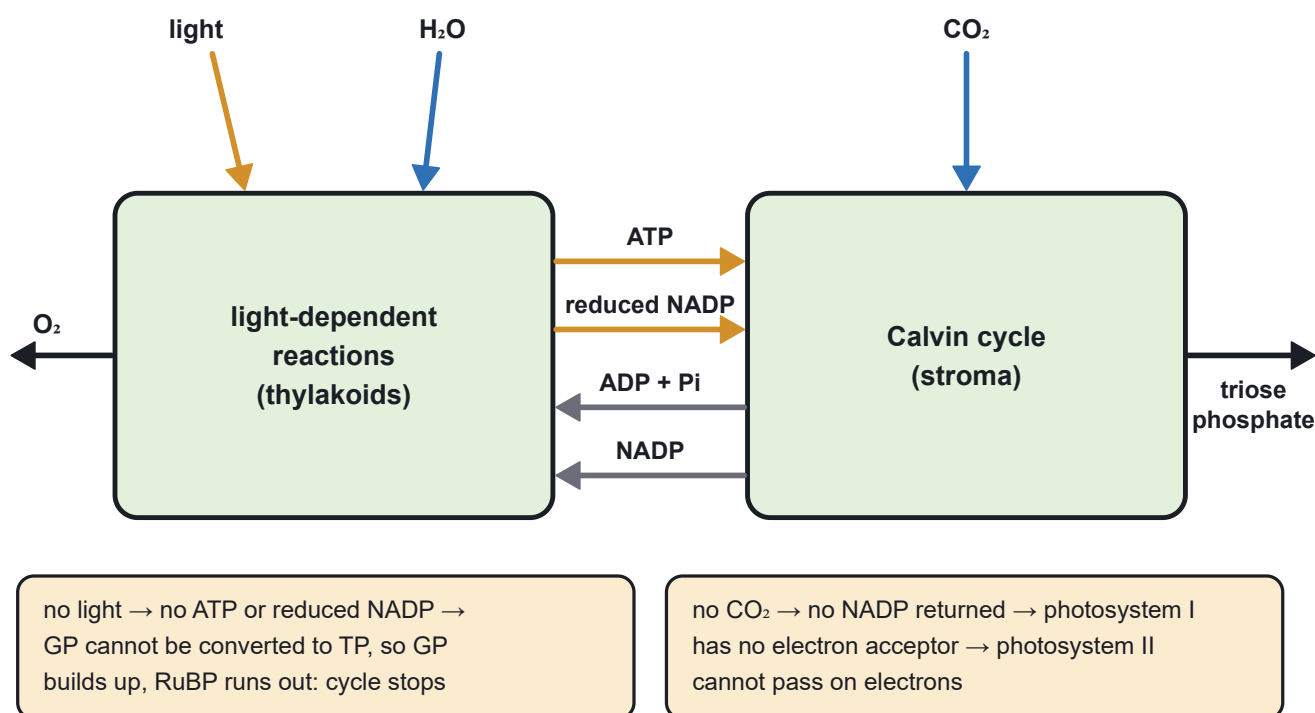
All of the carbon in compounds in photosynthesising organisms is fixed in the Calvin cycle. Glucose is only one product. Other carbon compounds are made by metabolic pathways that can be traced back to an intermediate in the cycle, usually triose phosphate. You do not need the details of these pathways.

Carbon compounds made from Calvin cycle products		
Product	Made from	Mineral nutrients also needed
Glucose, sucrose, starch, cellulose	Triose phosphate	None
Fatty acids and glycerol (lipids)	Triose phosphate and other intermediates	None (phosphate for phospholipids)
Amino acids (proteins)	Calvin cycle intermediates	Nitrogen (nitrate or ammonium); sulfur for some
Nucleotides (DNA, RNA, ATP)	Calvin cycle intermediates	Nitrogen and phosphate

Interdependence of the light-dependent and light-independent reactions

The light-dependent reactions supply ATP and reduced NADP to the Calvin cycle; the Calvin cycle returns ADP, phosphate and NADP. Neither can continue for long without the other.

- **Lack of light** stops the light-dependent reactions, so no ATP or reduced NADP is made. GP cannot be converted to TP, so GP accumulates and RuBP is not regenerated. The Calvin cycle stops.
- **Lack of CO₂** stops carbon fixation, so ATP and reduced NADP are not used. NADP is not regenerated, so photosystem I has no electron acceptor and the electron carriers stay reduced. Electrons from photosystem II cannot be passed on, so **photosystem II stops functioning**, and photolysis and oxygen production stop.



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The two sets of reactions **depend on each other**: ATP and reduced NADP go one way, ADP, Pi and NADP come back.

Exam tip: For a change in conditions, predict what builds up and what runs out. Darkness: GP rises and RuBP falls. Low CO₂: RuBP rises and GP falls.

Quick check

1. Write the word equation for photosynthesis.
2. Where does the oxygen released by photosynthesis come from?
3. How is an R_f value calculated?
4. What is the difference between an absorption spectrum and an action spectrum?
5. **HL only** What are the substrates and product of the reaction catalysed by Rubisco?

Quick check answers

Photosynthesis

1. carbon dioxide + water $\rightarrow$ glucose + oxygen
2. The splitting of water.
3. Distance moved by the pigment $\div$ distance moved by the solvent front.
4. An absorption spectrum shows light absorbed by a pigment at each wavelength; an action spectrum shows the rate of photosynthesis at each wavelength.
5. Substrates: RuBP and CO₂. Product: glycerate 3-phosphate (GP).